Subterranean Biology 40: 109–128 (2021)
doi: 10.3897/subtbiol.40.77451
https://subtbiol.pensoft.net

RESEARCH ARTICLE

A peer-reviewed open-access journal
Subterranean Biology
Published by The International Society for Subterranean Biology

The first troglobitic *Pseudochthonius* Balzan, 1892 (Pseudoscorpiones, Chthoniidae) from the karst area of Serra do Ramalho, Brazil: a threatened species

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Academic editor: Martina Pavlek | Received 4 November 2021 | Accepted 8 November 2021 | Published 23 November 2021

http://zoobank.org/3209150E-9866-42B1-A8AA-42C5B67A9FEF

Citation: Assis LD, Schimonsky DMV, Bichuette ME (2021) The first troglobitic *Pseudochthonius* Balzan, 1892 (Pseudoscorpiones, Chthoniidae) from the karst area of Serra do Ramalho, Brazil: a threatened species. Subterranean Biology 40: 109–128. https://doi.org/10.3897/subtbiol.40.77451

Abstract
Pseudochthonius ramalho **sp. nov.** is described to Gruna do Vandercir cave, in the Serra do Ramalho karst area, southwestern Bahia, Brazil. This area has an extensive limestone outcrop, with several caves, and the occurrence of potential minerals that are financially attractive for mining projects. The new species shows troglomorphic characteristics such as the depigmentation of the carapace and absence or reduction of eyes. It is a rare troglobitic species, and following the criteria of IUCN, we categorized the species as Critically Endangered – CR, IUCN criteria B1ab(iii)+2ab(iii). According to Brazilian legislation, locations, where critically endangered species live, can be protected by law, and we consider this cave/region to be of maximal relevance for protection.

Keywords
Arachnida, Bahia State, Chthoniinae, conservation, endemic, troglobite

Introduction

Pseudoscorpions are represented in the Brazilian fauna by 14 families and 176 species (Harvey 2013; Schimonsky and Bichuette 2019a; Benavides et al. 2019; Viana and Ferreira

2020; Bedoya-Roqueme et al. 2021). Only 33 species are known to occur in subterranean habitats (caves) belonging to 12 genera and eight families (Beier 1969; Mahnert 2001; Andrade and Mahnert 2003; Ratton et al. 2012; Schimonsky et al. 2014; Viana et al. 2018; Schimonsky and Bichuette 2019b; Viana and Ferreira 2020; Bedoya-Roqueme et al. 2021). The family Chthoniidae includes three subfamilies, Chthoniinae, Tridenchthoniinae, and Lechtyiinae, currently with 50 genera and 769 described species in the world (Harvey 2013; Zhang and Zhang 2014; Gao et al. 2018; Zaragoza and Reboleira 2018; Benavides et al. 2019). It occurs in several regions of the world, with the greatest diversity in the USA (147), Italy (84), Spain (59), Australia (36) (Harvey 2013). In Brazil, the family Chthoniidae has 29 species in eleven genera, *Heterolophus* Tömösváry, 1884, *Tridenchthonius* Balzan, 1887, *Lechthyia* Balzan, 1892, *Pseudochthonius* Balzan, 1892, *Compsaditha* Chamberlin, 1929, *Tyrannochthonius* Chamberlin, 1929, *Austrochthonius* Chamberlin, 1929, *Sororditha* Chamberlin & Chamberlin, 1945, *Neoditha* Feio, 1945, *Cryptoditha* Chamberlin & Chamberlin, 1945 and *Lagynochthonius* Beier, 1951 (Harvey 2013; Lira et al. 2020).

The genus *Pseudochthonius* is characterized by the absence of an intercoxal tubercle, the presence of coxal spines on coxae I and II, and in most cases, having strongly sigmoid palpal chelal fingers (Muchmore 1986; Mahnert and Adis 2002). *Pseudochthonius* is known from Asia (one species from Saudi Arabia), Africa (five species distributed in the Republic of Congo and Ivory Coast), Central America [six living species of which two are troglobites: *P. troglobius* Muchmore, 1986 from Mexico (Muchmore 1986) and *P. arubensis* Wagenaar-Hummelinck, 1948, from the Netherlands Antilles (Wagenaar-Hummelinck1948) and one fossil species (Schawaller 1980)] and South America, with nine species in Brazil (Harvey 2013; Mahnert et al. 2014; Lira et al. 2020), followed by Ecuador with three, Venezuela with two and Uruguay with one species (Harvey 2013). In Brazil, two *Pseudochthonius* species are troglobites (Mahnert 2001), i.e., they have exclusive populations in the hypogean environment: *P. strinatii* Beier, 1969 and *P. biseriatus* Mahnert, 2001. *Pseudochthonius* is one of the four Chthoniidae genera that occurs in Brazilian caves, previously reported only for13 caves, with four species (Beier 1969; Mahnert 2001). Recently, this genus was recorded in other karst areas and biogeographical provinces, increasing its distribution to 37 caves (Schimonsky and Bichuette 2019b).

Here, we describe a new *Pseudochthonius* species found in a single cave in the Serra do Ramalho karst area, southwestern Bahia, Brazil. We also provide data regarding the conservation status of the species and the area.

Material and methods

Study area

The karst area of the Serra do Ramalho region (Fig. 1), southwestern Bahia, Brazil, is composed of rocks containing rare metals with mining potential (Silva Junior and Campos 2016). The region is formed by extensive limestone areas with many caves

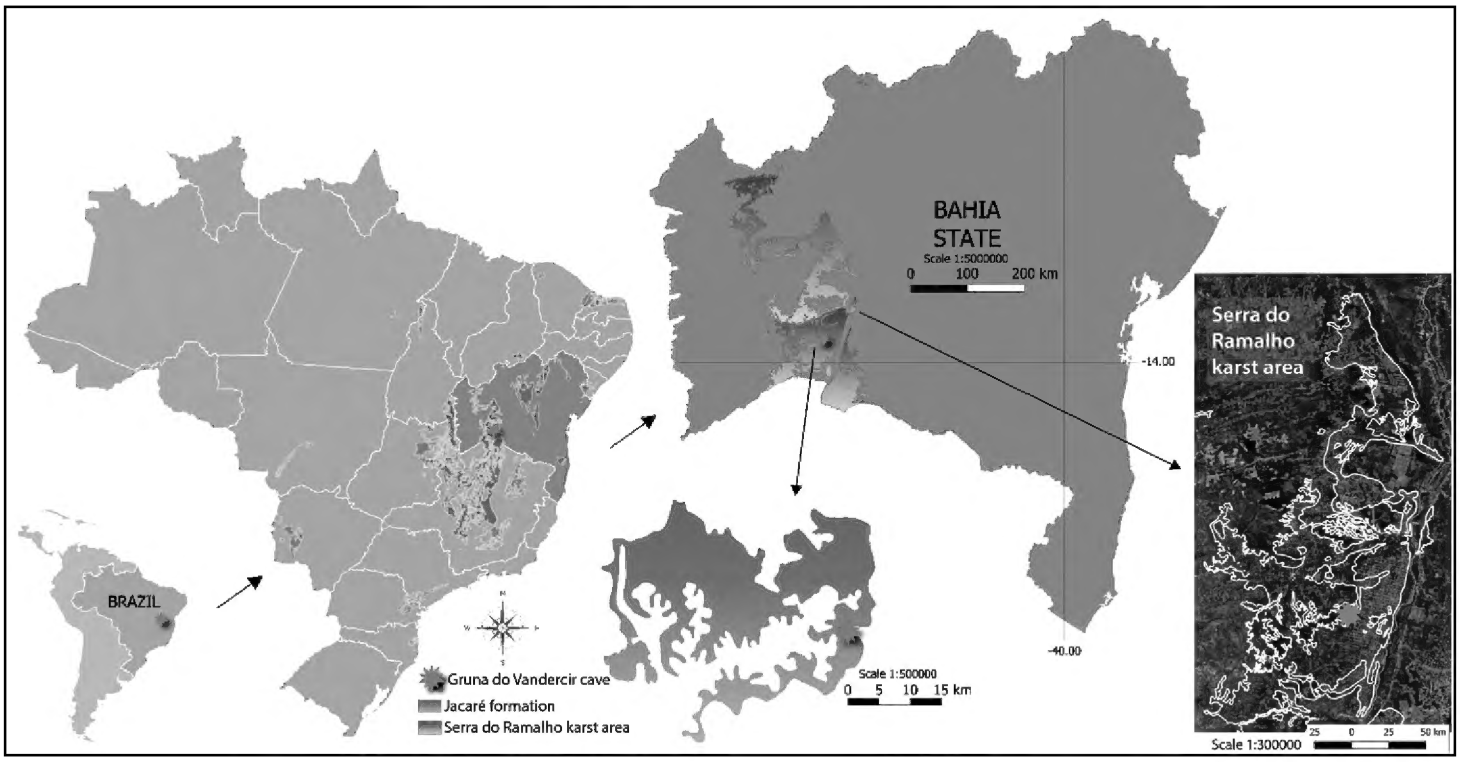

Figure 1. Map depicting the known distribution of *Pseudochthonius ramalho* sp. nov. in Gruna do Vandercir cave, located in Bahia state. The cave belongs to karst area in Jacaré formation, Bambuí group.

and karst system formations (Fig. 2), sheltering a high diversity (e.g., Bichuette and Trajano 2004, 2005; Bichuette and Rizzato 2012; Gallão and Bichuette 2018). It belongs to the Jacaré formation, Bambuí group, composed of dark, heterogeneous limestones, presenting intercalations with layers of claystone, deposited about 750 to 600 million years ago (Rubbioli et al. 2019). The region located in the middle of the São Francisco River basin is characterized by a tropical dry climate, with a dry winter and an average annual precipitation rate between 800 and 1000 mm (Gonçalves et al. 2018).

Material examined and treatment of specimens

Specimens were prepared by immersion in 85% lactic acid at room temperature for two weeks (Judson 1992). They were then examined by preparing temporary slide mounts with 10 mm coverslips supported by sections of nylon fish line (Harvey 2021). Specimens were examined with Nikon SMZ660 Stereomicroscope and Leica DMLS compound microscope and the male holotype was illustrated with the aid of a camera lucida. The female specimen was examined and illustrated through a Scanning Electron Microscope (SEM, FEI Quanta 250) in low-vacuum mode (ESEM) located at the "Instituto Nacional de Ciência e Tecnologia dos Hymenoptera Parasitoides da Região Sudeste Brasileira". After the study, the male specimen was cleaned in water and returned to 70% ethanol with its dissected parts in glass vials, and the female specimen was stored dry, due to the SEM picturing process.

Images (Figs 4, 10) were taken with a Leica DFC 295 camera attached to a Leica M205C stereomicroscope with a Planapo 1.0× objective. Figures were produced from stacks of images on Leica Application Suite (LAS) software v3.7. The drawings were

Figure 2. A Gruna do Vandercir cave **B** surroundings of Gruna do Vandercir cave with its dry characteristic vegetation (Images **A** Adriano Gambarini **B** Maria Elina Bichuette).

digitized and vectorized on Illustrator CC 2019. The maps were produced with the software QuantumGis Desktop 3.6.0 (QGis Open Source Geospatial Foundation). The coordinates were obtained from field trips to the study location with a global positioning system (GPSGarmin 60CSx).

The examined specimens are deposited in Laboratório de Estudos Subterrâneos, in Universidade Federal de São Carlos (LES, curator: Maria Elina Bichuette). For comparative purpose of some morphological characters like classical troglomorphisms in pseudoscorpions (eyes/ocular structures, proportionally longer body, and ratio pedipalpal chela/carapace), the new species was compared to two hypogean species, and one undetermined epigean species of *Pseudochthonius* sp.

Comparative material. Brazil – **Parana Forest Province** • *Pseudochthonius strinatii*;1♂, São Paulo, Iporanga, Parque Estadual Turístico do Alto Ribeira, Sumidouro da Passoca cave; 24°33'57"S, 48°43'W; 03.xii.2013; Bichuette ME, Gallão JE, Fernandes CS, Rizzato PP, Fonseca R and Arnone I leg.; LES9391. – **Parana Forest Province** • *Pseudochthonius biseriatus*; 1♂Minas Gerais, Itacarambi, Olhos d'Água cave; 15°7'0.10"S, 44°10'0.10"W; 24.vii.2012; Bichuette ME, Gallão JE, and Rizzato PP leg.; LES9434. – **Caatinga Province** • *Pseudochthonius* sp. undetermined species; 1♂; Bahia, Carinhanha, epigean habitat near Viração cave; 26.vii.2012; LES9629.

Terminology and mensuration

The terminology and measurements mostly follow Chamberlin (1931). Legs, pedipalps, and trichobothria terminology follows Harvey (1992) except for the chelal movable finger, which follows Mahnert et al. 2014. For chelicera Judson (2007), chaetotactic formulae of chelicera follow Gabbutt and Vachon (1963) and the duplex trichobothria follow Judson (2018).

Abbreviations

♂	male;	♀	female.

chelal trichobothria:

b	basal;	***isb***	interior sub–basal;	***est***	exterior sub–terminal;
sb	sub–basal;	***ist***	interior sub–terminal;	***et***	exterior terminal;
st	sub–terminal;	***it***	interior terminal;	***dx***	duplicate trichobothria.
t	terminal;	***eb***	exterior basal;		
ib	interior basal;	***esb***	exterior sub–basal;		

cheliceral setae:

gl	galeal;	***db***	dorsal basal;	***di***	isolated subapical tooth.
dt	dorsal terminal;	***vt***	ventral terminal;		
dst	dorsal sub–terminal;	***vb***	ventral basal;		

The following body structures were measured and compared proportionally for the characterization of possible troglomorphisms: carapace; chelal hand; chelal fixed finger (Christiansen 2012; Harvey and Wynne 2014; Feng et al. 2020; Harvey and Cullen 2020).

Results

Taxonomy

Family Chthoniidae Daday, 1889
Subfamily Chthoniinae Daday, 1889
Tribe Chthoniini Daday, 1889
Genus *Pseudochthonius* Balzan, 1892

Pseudochthonius ramalho **Assis, Schimonsky & Bichuette, sp. nov.**
http://zoobank.org/5558E734-180A-43B0-B4D4-5BDA609AC030
Figs 3–8

Type material. ***Holotype*****:** 1 ♂ (LES9601) Brazil Caatinga province, Serra do Ramalho karst area, Serra do Ramalho, Bahia, Gruna do Vandercir cave; 13°38'11.40"S, 43°50'5.10"W; 31 May 2012; Bichuette ME, Gallão JE, Hattori N leg. ***Paratype*****:** 1 ♀ (LES9602), same data as holotype.

Etymology. The species is named after the region of Serra do Ramalho due to its importance regarding the speleological heritage and the unique fauna and flora diversity. The name is to be treated as a noun in apposition.

Figure 3. Holotype of *Pseudochthonius ramalho* sp. nov. in natural habitat, at Gruna do Vandercir cave, Serra do Ramalho, Bahia. (Image: Adriano Gambarini).

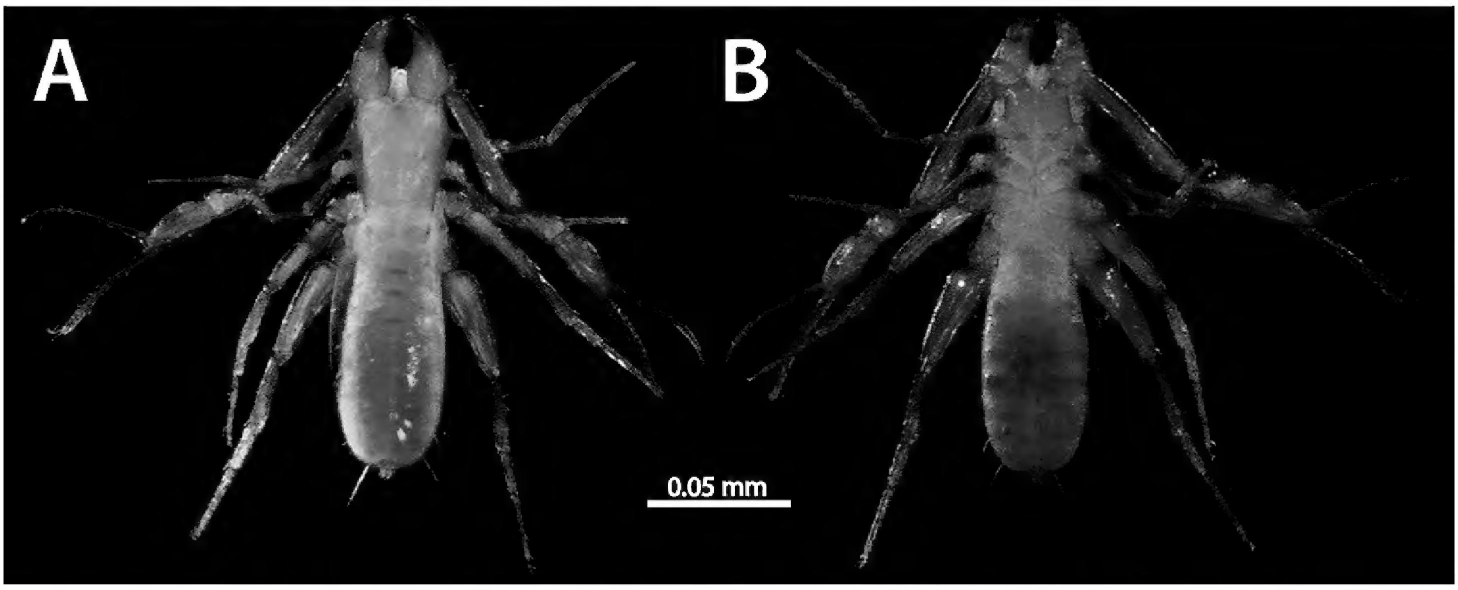

Figure 4. *Pseudochthonius ramalho* sp. nov. holotype male, habitus **A** dorsal view **B** ventral view.

Diagnosis. *Pseudochthonius ramalho* sp. nov. can be identified by the following combination of characters: eyes absent (♂) or with eyes-spots (♀); the middle and distal fixed chelal finger teeth positioned two by two with 29–30 acuminate teeth and 3–4 slightly basally rounded ones, the presence of two rounded micro–denticles, along with pedipalpal fixed finger teeth in males; trichobothria *ist* closer to *esb* than to the *est* (ratio *ist-est*/*ist-esb* = 4.71); serrula exterior with 13 (♂) or 14 (♀) lamellae, rallum with seven blades, and coxae I and II with 3 to 5 coxal spines.

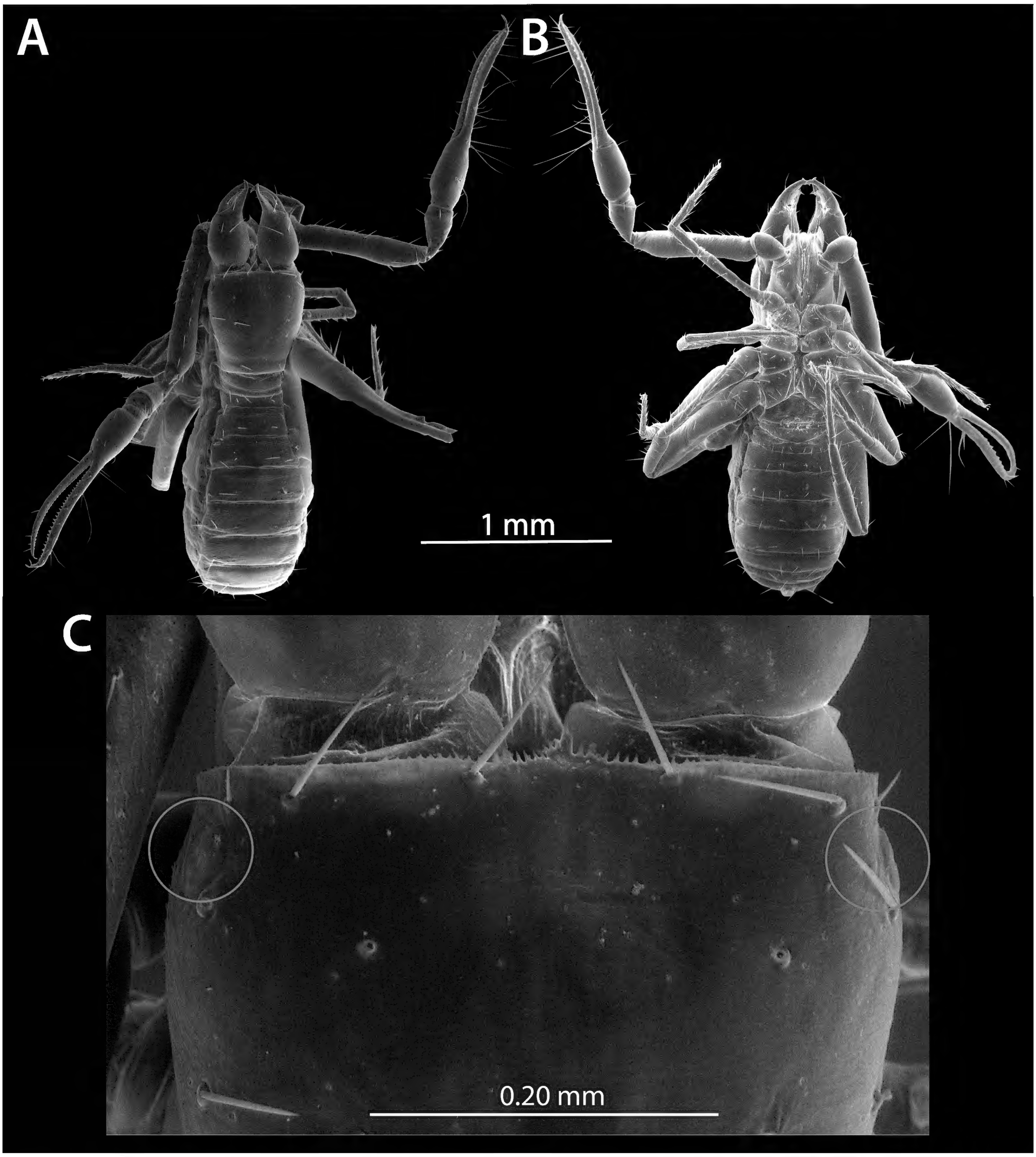

Figure 5. *Pseudochthonius ramalho* sp. nov. scanning electron images. Paratype female, habitus **A** dorsal view **B** ventral view **C** zoom in on the anterior margin of the carapace with eye spots denoted with red circle. (Images: Luciana B. R. Fernandes).

Description (adult ♂ and ♀). ***Body*:** Coloration of specimens in 70% ethanol yellowish brown and translucent pedipalps, tergites III–V with a dark median mark, and a darker abdominal region. Live specimens present a light pinkish color on their carapace and appendages, and a light brown abdomen. Female is slightly smaller than male.

Chelicera (Figs 6A, 7B, C): five setae on left hand, with one seta on the basal position of the fixed finger and one nearly the basal seta on the movable finger; six setae on the right hand; without the lateral microsetae; fixed finger with 10–11 (♂

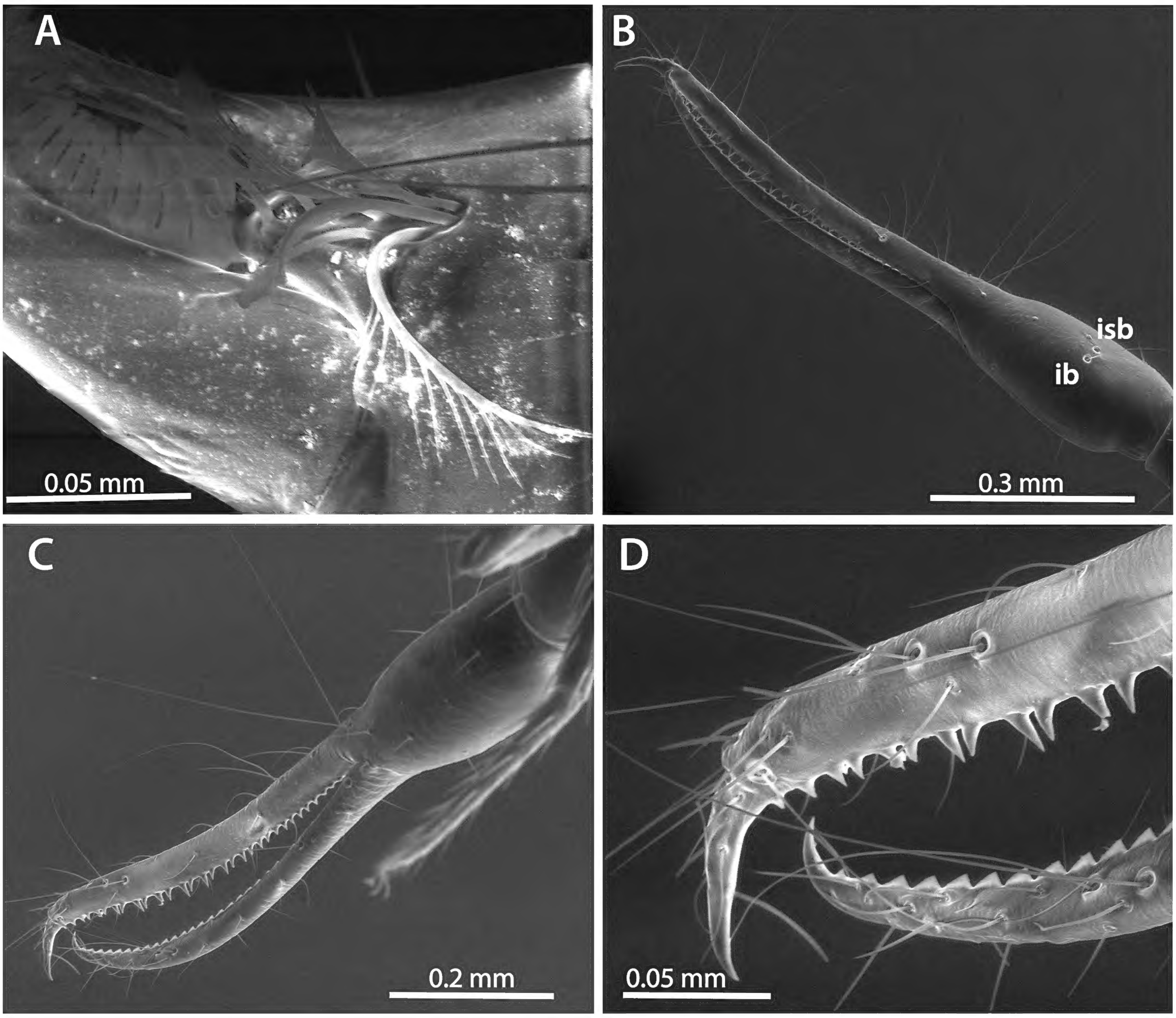

Figure 6. *Pseudochthonius ramalho* sp. nov. female paratype scanning electron images **A** detail on the right chelicera of serrula exterior and *rallum* **B** detail on the right pedipalp trichobothrium *isb* and *ib*, lateral **C** left pedipalp **D** detail on the left pedipalp teeth. (Images: L. B. R. Fernandes).

and ♀) teeth proximally reduced in size; movable finger with 9 (♂) or 8(♀) teeth proximally reduced in size, three distal teeth distinctly larger than others and with subapical isolated tooth (*di*). Spinneret moderately prominent and apically rounded in female, vestigial in male. Seta *gl* 0.15 mm from base of movable finger. Serrula exterior with 13 (♂),14 (♀) lamellae. Rallum with seven blades pectinated. Dorsal face of cheliceral palm with four lyrifissures, three lyrifissures situated near seta *dt* and one situated posteriorly.

Pedipalp (Figs 6B–D, 8A–F): 1.4 (♂),1.2 (♀) × longer than carapace and 2.2 (♂), 2.6 (♀) × longer than patella; movable finger 1.6 (♂), 1.9 (♀) × longer than hand; fixed finger 1.65 (♂),1.72 (♀) × longer than hand. Fixed chelal finger long and strongly sigmoid in its distal half. Male fixed finger with 33 acute teeth, distinctly separated from each other, but paired and in each pair, one tooth is slightly directed to inside and the other to the outside, and micro–denticles in two interdental spaces, on teeth 15 and 29, respectively. Female fixed finger with 31 teeth arranged as in as in male. Mov-

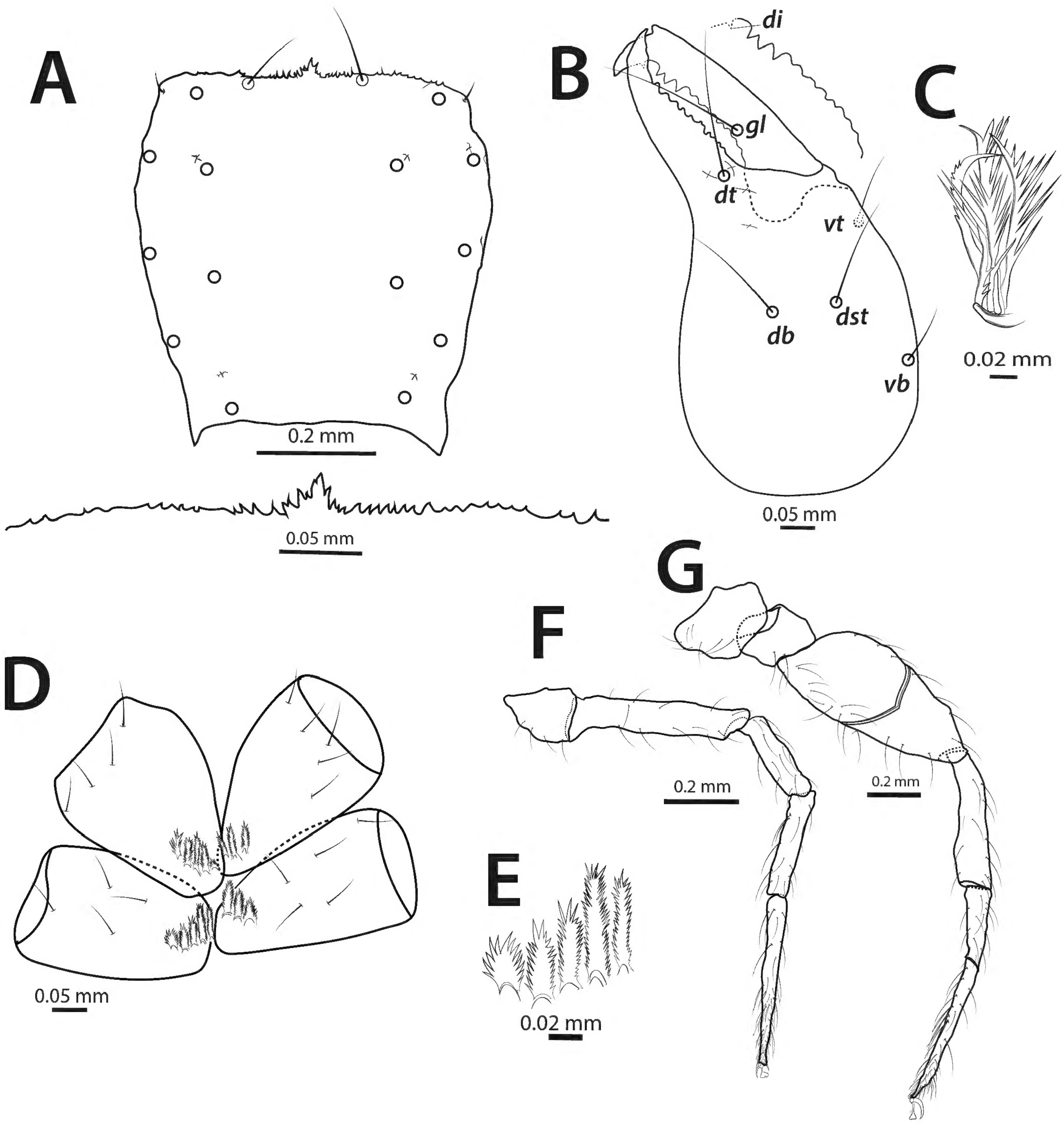

Figure 7. *Pseudochthonius ramalho* sp. nov., male **A** carapace dorsal view, and detail of the anterior margin (with the epistome) **B** right chelicera (dorsal view) **C** detail of the rallum **D** coxa I and II **E** details of coxal spines **F** leg I (lateral view) **G** leg IV (lateral view).

able finger with 30–33(♂ and ♀) flattened and separated teeth. Trichobothria: *ib* and *isb* situated close to each other sub–medially in the dorsal region of the chelal hand; *eb* closer to *esb* than to *ist*, forming a straight oblique row at the base of the fixed chelal finger; *ist* closer to *esb* than to the *est* (ratio *ist–est/ist–esb* = 4.71); *et* slightly near the tip of the fixed finger, near to the chelal teeth; *dx*, located near to the end of the fixed finger; *sb* closer to *b* than to *st* in the movable chelal finger (ratio *sb–st/sb–b* = 3.37); *t* closer to *st* and situated at the same level as *est*.

Carapace (Fig. 7A): Carapace 1.09× longer than broad, posteriorly constricted, chaetotaxy 4:4:4:2:2 (16), one preocular microseta on each side; eyes absent on male

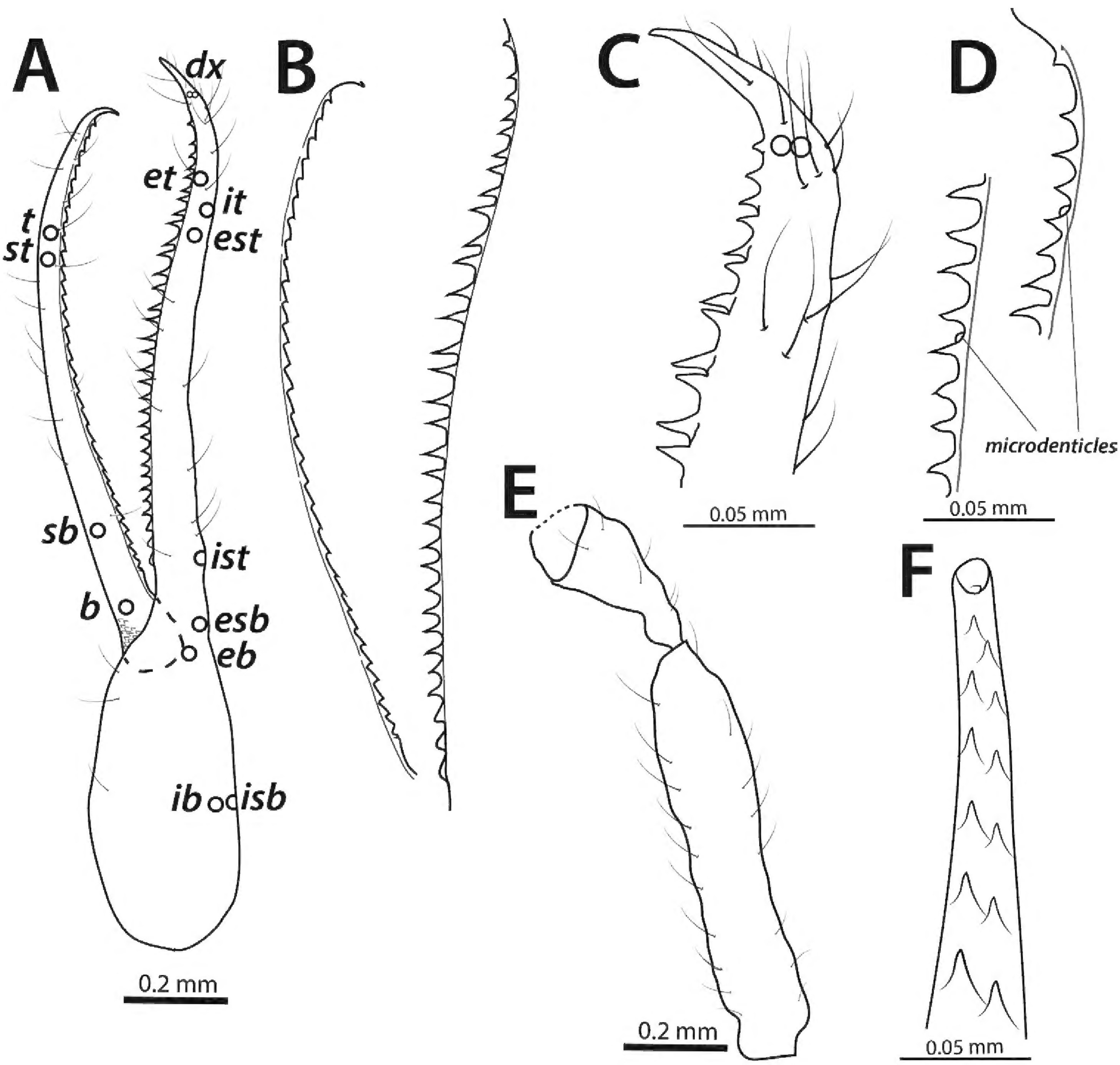

Figure 8. A holotype left pedipalp showing the trichobothria distribution **B** details of chelal teeth **C** distal part of fixed chelal finger (lateral view) **D** detail with emphasis on the micro–denticles in two interdental spaces, on teeth 15 and 29, respectively **E** pedipalp femur **F** distal part of fixed chelal finger (ventral view).

and a tiny eyespot on female; anterior margin distinctly serrate with median denticles larger than lateral ones; epistome prominent and dentate (Fig. 7A–C); 3 lyrifissures anteriorly, 1 medially and 2 posteriorly.

***Abdomen*:** Chaetotaxy of tergites I–XI: ♂, 4: 4: 4: 4: 6: 6: 6: 6: 6: 5: 3; ♀, 4: 4: 4: 4: 5: 6: 6: 6: 6: 5: 3. Chaetotaxy of sternites III–XI: (♂/♀) 12: 13: 8: 8: 8: 8: 6: 5: 2, anal cone 0/2 setae.

***Genital area*:** Anterior genital operculum with 8 (♂), 9 (♀) marginal and discal setae, arranged triangularly in male, with 7–8 unmodified marginal setae on each side; posterior operculum with 6 setae in female.

Coxae (Fig. 7D, E): Manducatory process distally acute, with 2 setae; pedipalpal coxa with 3 setae, coxa I and II with 4–5 setae on anterior margin and 3–5 highly dented coxal spines in decreasing size distally, coxa III with 7 setae and coxa IV with 8 setae; intercoxal tubercle absent.

Table 1. Measurements (in mm) and proportions (l/b, length/breadth; l/d, length/depth) of the holotype male and paratype female of *Pseudochthonius ramalho* sp. nov.

	Holotype (Paratype)	Holotype (Paratype) l/b; l/d
Body	1.55 (1.45)	
Carapace	0.46/0.42 (0.45/0.42) narrower part posteriorly 0.27 (0.275)	1.1 (1.0)
Pedipalpal trochanter	0.18/0.12 (0.16/0.11)	1.6 (1.5) l/b
Pedipalpal femur	0.65/0.16 (0.54/0.13)	4.1 (4.1) l/b
Pedipalpal patella	0.29/0.15(0.25/0.12)	4.1 (2.1) l/b
Pedipalpal chela	0.90/0.20 (0.81/0.14)	4.5 (5.8) l/d
Pedipalpal hand	0.35/0.20 (0.29/0.14)	1.9 (2.1) l/d
Pedipalpal fixed finger	0.58/0.05 (0.50/0.02)	
Pedipalpal movable finger	0.57/0.04 (0.55/0.02)	
Chelicera	0.39/0.19 (0.38/0.18)	2.1 (2.1) l/b
Chelicera movable finger	0.18 (0.17)	
Leg I femur	0.35/0.06 (0.36/0.05)	5.9 (7.2) l/d
Leg I patella	0.18/0.05 (0.16/0.05)	3.6 (3.2) l/d
Leg I tibia	0.21/0.04 (0.15/0.04)	5.2 (3.8) l/d
Leg I tarsus	0.33/0.03 (0.29/0.03)	11 (9.6) l/d
Leg IV trochanter	0.18/0.16 (0.16/0.13)	1.1 (1.2) l/b
Leg IV femur + patella	0.76/0.28 (0.70/0.12)	2.7 (5.8) l/d
Leg IV tibia	0.38/0.08 (0.16/ 0.05)	4.7 (3.2) l/d
Leg IV basitarsus	0.19/0.05 (0.18/0.05)	3.8 (3.6) l/d
Leg IV telotarsus	0.35/0.03 (0.34/0.02)	11.6 (17) l/d

Legs (Fig. 7F, G): Typical of the genus (Chamberlin 1929).
***Measurements and ratios*:** see Table 1.

Taxonomic remarks

The new species *Pseudochthonius ramalho* sp. nov. is compared with other hypogean and epigean *Pseudochthonius* species. It most resembles other Brazilian species of *Pseudochthonius* that lack eyes, and occur in caves, like *P. strinatii* and *P. biseriatus*. *Pseudochthonius ramalho* sp. nov. has 5 coxal spines, almost ever–increasingly arranged; sternites V–VIII with 8 setae on sternal chaetotaxy; trichobothrium *ist* is 4× farther from *est* than from *esb*; the middle and distal fixed chelal finger teeth differ only in their direction, but not in their size or shape, with teeth arranged two by two; male is slightly larger; it has pedipalpal patella and pedipalpal femur proportionally larger and smaller (4.1 ♂ and ♀). Differently, *Pseudochthonius strinatii* has 2 longer and 2 shorter coxal spines; sternal chaetotaxy with 6 setae on each sternite; the position of trichobothria *ist* is 3× farther from *est* than from *esb*; its fixed chelal finger teeth show heterodontism; pedipalpal patella and pedipalpal femur proportionally smaller (2.0 ♂) and larger (5.3–6.1 ♂), respectively. *Pseudochthonius biseriatus* has 2 setae on tergites I and II, a rallum with nine setae, and 37–41 teeth arranged in an offset manner; chelal length 1.24–1.39. In contrast, *P. ramalho* sp. nov. has four setae on tergites I and II, a rallum with seven blades, and chela fixed finger with 30–33 teeth; chelal length 0.81–0.90. All three species share the unpigmented tegument with other troglobitc

Pseudochthonius species, like *P. troglobius* and *P. pulchellus* (Ellingsen, 1902). However, *P. ramalho* sp. nov. differs from these and from *P. biseriatus* and *P. strinatii* due the presence of ocular spots in the female. Other nontroglobitic *Pseudochthonius* present in Brazilian caves, have eyespots (*P. gracilimanus* Mahnert, 2001 and *P. ricardoi* Mahnert, 2001). *Pseudochthonius troglobius* has a pedipalpal fixed finger with 65 teeth, and proportionally larger body features (e.g., movable finger 2.14× longer than hand). This is different from *P. ramalho* sp. nov. with 30–33 teeth in the fixed pedipalpal finger and a proportionally smaller body (e.g., movable finger 1.6× longer than hand). Considering the number of marginal teeth on the pedipalpal movable finger, *P. ramalho* sp. nov. resembles *P. gracilimanus* and *P. strinatii* with 30–33 teeth (Beier 1969; Mahnert 2001), but it differs from the *P. biseriatus* (34–37), *P. ricardoi* (43), and the epigean *P. orthodactylus* Muchmore, 1970 (7) (Muchmore 1970; Mahnert 2001). Also, *Pseudochthonius ramalho* sp. nov. tarsus of leg I (9.6–11× longer than deep) is similar to other brazilian cave-dwelling species like *P. biseriatus* (10.3–11.0), *P. strinatii* (9.5–10.7), *P. ricardoi* (10.1) and is longer than the epigean species *P. tuxeni* (7.3) (Beier 1969; Mahnert 1979; Mahnert 2001).

Discussion

Distribution of the genus *Pseudochthonius* in Brazil

Species of *Pseudochthonius* occur in five Brazilian states (Fig. 9): in the state of São Paulo (southeastern Brazil) with representatives of *P. strinatii* and *P. ricardoi* in cave habitats (Alto Ribeira karst area) and *P. brasiliensis* (in the region of Barueri); in state of Minas Gerais (southeastern Brazil), with the troglobitic species *P. biseriatus* endemic to the cave Olhos d'Água; in the state of Bahia (northeastern Brazil), with the new species described herein *P. ramalho* sp. nov., troglobitic and endemic of to Gruna do Vandercir cave, and also *P. gracilimanus* in cave habitat; in the state of Pará (northern Brazil) with representatives *P. orthodactylus* and *P. tuxeni*; and in the state of Amazonas (northern Brazil) the species *P. homodentatus* has been found in the Ducke Reserve and *P. heterodentatus* Hoff, 1946 was registered in the Urucu river basin (Aguiar and Bührnheim 1994). However, recently, this genus was recorded in other karst areas and biogeographical provinces, increasing its distribution to 37 more caves (Schimonsky and Bichuette 2019b).

Troglomorphic traits

Troglomorphic traits are characteristics that propose a relationship between hypogean species and the subterranean environment, associated with behavior, physiology, and mainly, morphology. Although these characteristics are useful to differentiate hypogean from epigean species, they do not explain the direct connection between the subterranean habitats and the species that inhabit it (Juberthie and Decu 1994).

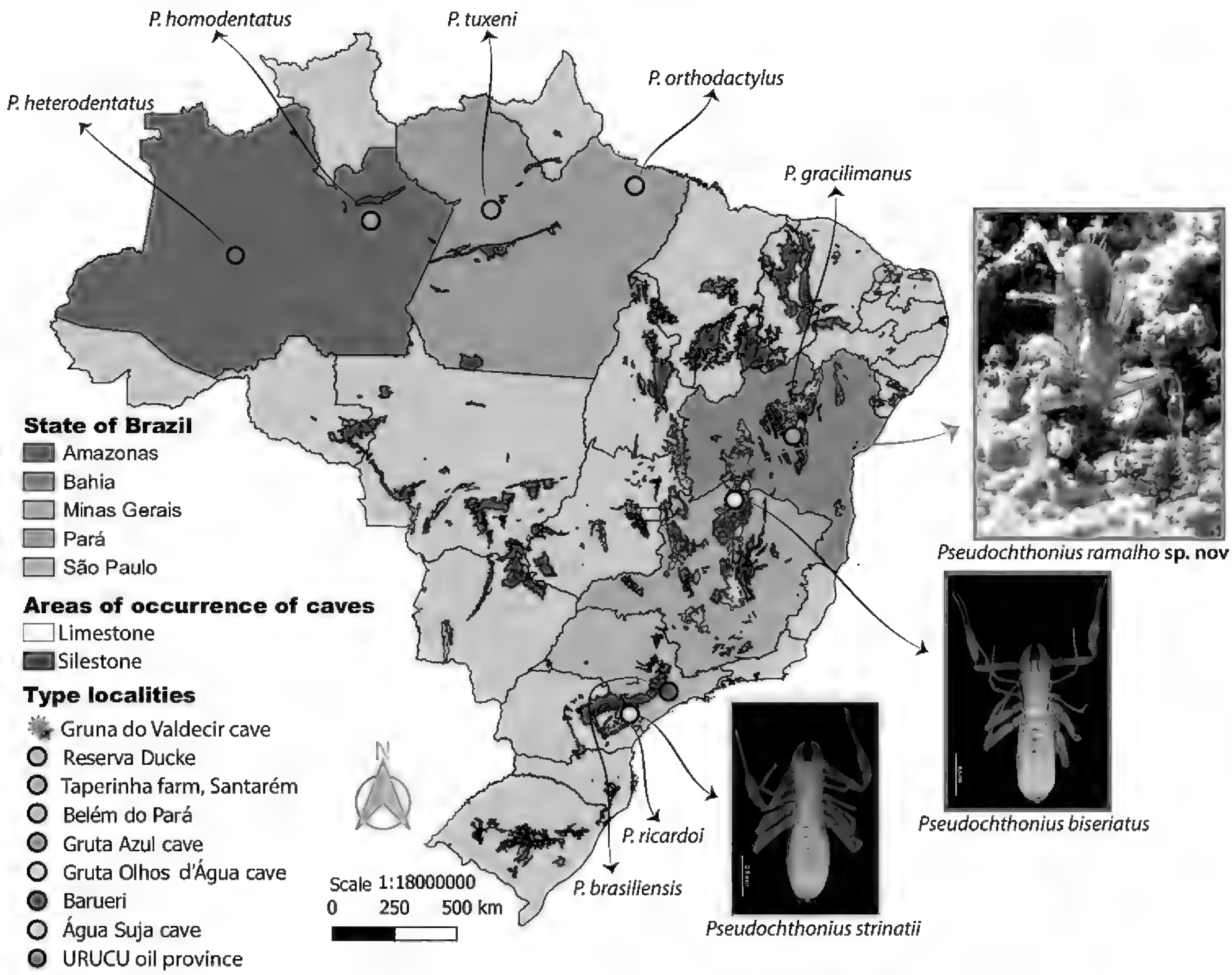

Figure 9. Distribution of epigean and hypogean *Pseudochthonius* species in Brazil, with troglobitic representatives detached.

Most families of pseudoscorpions have at least one troglomorphic feature. Chthoniidae can be considered one of the most important families regarding occurrence in subterranean habitats (Harvey et al. 2000; Reddell 2012). The most common troglomorphic characteristics are eye reduction and cutaneous melanin depigmentation, classified as regressive evolution (Christiansen 2012), in addition to progressive morphological changes, such as appendages elongation, which is fundamental for spatial orientation, defense, and predation in a habitat with the absence of light (Chamberlin and Malcolm 1960; Christiansen 2012). Examples of progressive morphological changes could be the comparison in the proportional length of the different body parts of different species, which highlights the appendages elongation, e.g., the tibia and the tarsus and of leg I. The hypogean species *Pseudochthonius ramalho* sp. nov. (5.2× longer than deep; 9.6–11× longer than deep), *P. biseriatus* (6.0× longer than deep; 10.3–11× longer than deep), *P. strinatii* (4.8× longer than deep; 9.5–10.7× longer than deep), *P. ricardoi* (5.3× longer than deep; 10.1× longer than deep). In the epigean species, *P. heterodentatus* (3.6× longer than deep; 10.0× longer than deep) and *P. tuxeni* (3.6× longer than deep; 7.3× longer than deep). Nevertheless, the epigean fauna of pseudoscorpions in South America is still little known (Mahnert and Addis 2002), which makes it

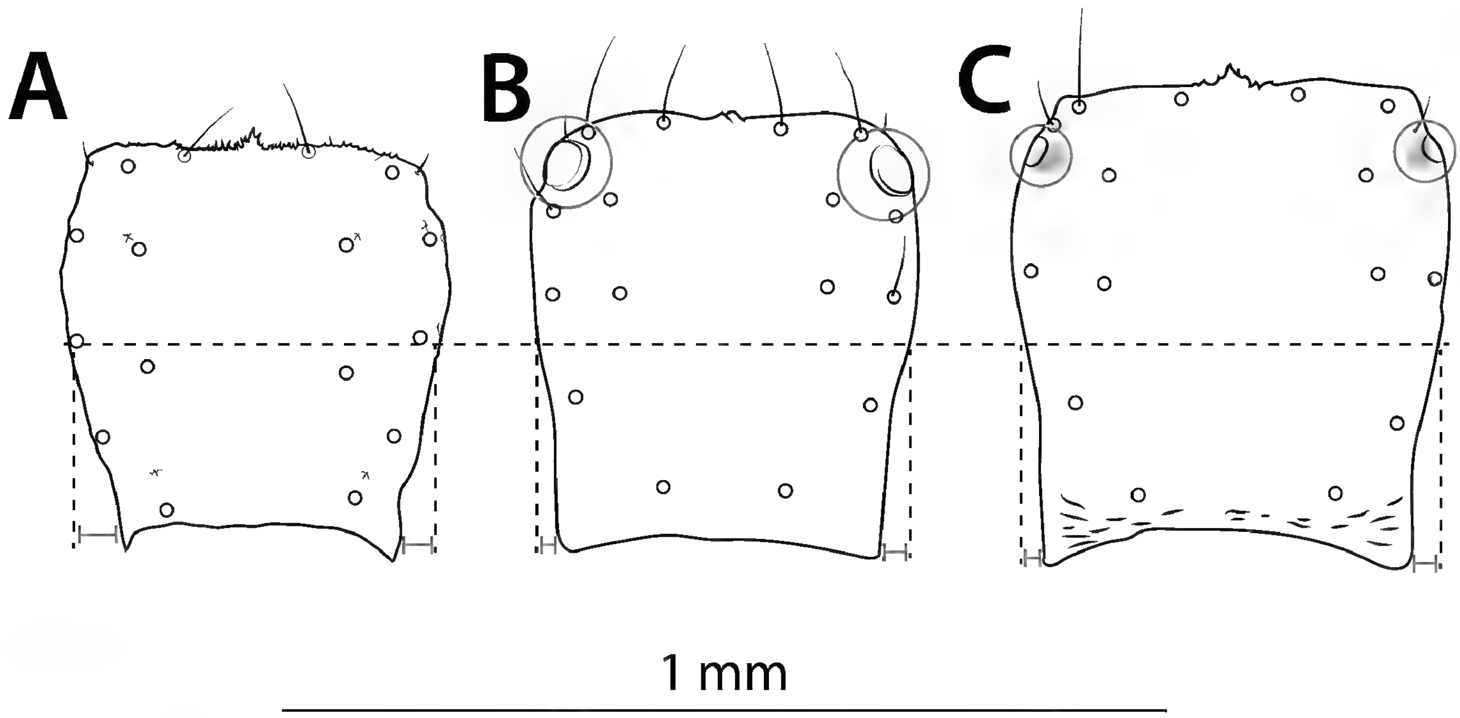

Figure 10. Morphological differences on the carapace of hypogean and epigean species of *Pseudochthonius*: eyes (denoted with red circle), and the narrowing of the posterior region of the carapace (marked with dashed line on the sides of the carapace) **A** hypogean *P. ramalho* sp. nov. (male) **B** epigean *P. thibaudi* **C** epigean *P. arabicus*.

difficult to compare hypogean and epigean individuals for the establishment of new troglomorphic characteristics. The female of the new species *P. ramalho* sp. nov. has ocular spots (Fig. 5) and the male has no ocular features. These characteristics should indicate a troglomorphic traits, that is, characters adapted to life in the subterranean environment (Fong 2012). Thus, these features can be compared with other Brazilian cave species (non–troglobitic), which have two small eyes (*P. gracilimanus*) or indistinct eye spots (*P. ricardoi*). When compared with some epigean species (*P. thibaudi* Castri, 1983 and *P. arabicus* Mahnert, 2014) it is noted that the absence or reduction of ocular traces is a troglomorphic trait (Fig. 10).

Pseudochthonius ramalho sp. nov. (Fig. 11A), *P. strinatii* (Fig. 11B) and *P. biseriatus* (Fig. 11C) show a narrowing in the carapace from the anterior to the posterior margin of the carapace of approximately 0.28 mm (anterior region 1.57× broad than longer), 0.26 mm (anterior region 1.61× broad than longer) and 0.29 mm (anterior region 1.37× broad than longer), respectively, when compared to the epigean *Pseudochthonius* sp. (unidentified species) (Fig. 11D), which is about 0.41 mm wide (anterior region 0.92× broad than longer), this could be another troglomorphic trait, like the so–called "false physogastry" in some cave beatles, like Leptodirini (Faille 2019).

In the pedipalpal chela of the three hypogean species, there is a slight decrease in the length and width of the hand and a significant increase in the length of the fixed finger. These pseudoscorpions have, respectively, the following length and width: hand (in mm) – 0.29/0.14 in *P. ramalho* sp. nov. (Fig. 11A1), 0.27/0.14 in *P. strinatii* (Fig. 11B1) and 0.26/0.13 in *P. biseriatus* (Fig. 11C1), and fixed finger (in mm) –0.59/0.04, 0.63/0.03 and 0.54/0.03, respectively. These values contrast with the ones observed in the epigean species *Pseudochthonius* sp.: hand (in mm) –0.31/0.19 and fixed finger

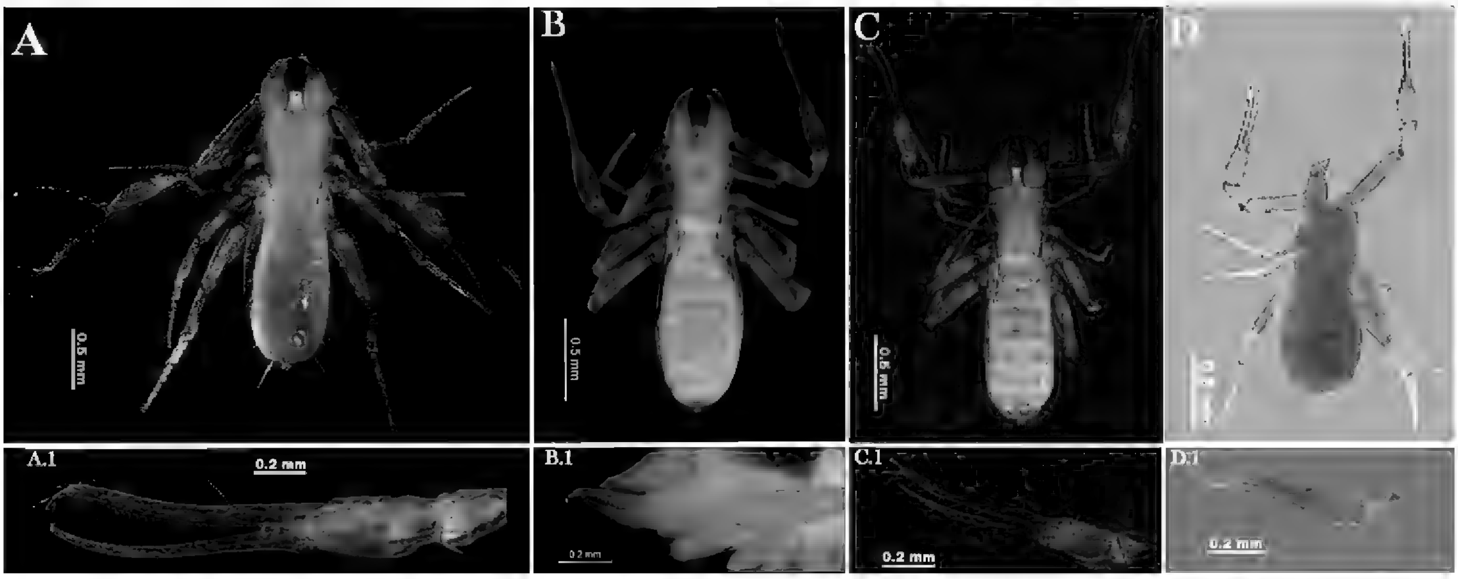

Figure 11. Comparison of morphology among some species of *Pseudochthonius* from Brazil **A** Holotype *Pseudochthonius ramalho* sp. nov. (troglobitic) (LES9601) and left chela (**A1**) **B** *Pseudochthonius strinatii* (troglobitic) (LES9391) and pedipalp detail (**B1**) **C** *Pseudochthonius biseriatus* (troglobitic) (LES9434) and pedipalp detail (**C1**) **D** *Pseudochthonius* sp. (epigean) (LES9629) and pedipalp detail (**D1**) (Images: **A** D. M. von Schimonsky; **A1–C1** L.B.R Fernandes; **D–D1** M. E. Bichuette).

(in mm) –0.49/0.04. Our observations corroborate that, for Chthoniidae, we cannot infer troglomorphism concerning to a single character (such as body pigmentation), but rather to a combination of traits (eyes/ocular structures, thinning of the cuticle, proportionally longer body, pigmentation, and ratio pedipalpal femur/carapace).

Conservation remarks

The Serra do Ramalho region is formed by several masses of carbonate rocks, thus enabling the occurrence of many karst features, including caves. Cave extensions range from hundreds of meters to more than 5 km, some exceeding 15 km (Rubbioli et al 2019). Another record of great importance is the great potential regarding subterranean fauna for both invertebrates and vertebrates (e.g., Baptista and Giupponi 2002; Pérez and Kury 2002; Bichuette and Trajano 2004; Bichuette and Trajano 2005; Trajano et al. 2009; Bichuette and Rizzato 2012). The issues related to the preservation of the subterranean environments in this karst region are directly influenced by the corresponding epigean environment. As the subterranean organisms use allochthonous organic matter, they rapidly suffer from the effects of any changes that occur to the epigean environment, e.g., deforestation and surface water pollution. Thus, studies demonstrate that the vast diversity of subterranean fauna is extremely important as an indicator of the health of the overall area (Bichuette et al. 2013; Gallão and Bichuette 2018). However, the Serra do Ramalho region is not yet inserted in any conservation units (e.g., State Park) and it is exposed to risks such as deforestation (e.g., wood for charcoal production), agriculture and mining projects due to the presence of rare metals (e.g., niobium) (Silva Junior and Campos 2016; Gallão and Bichuette 2018). The exploration and extraction of these metals can destroy entire caves and systems, leading to the extinction of isolated populations in these habitats (Culver 1986). This, coupled

with the lack of laws that effectively protect caves in Brazil, leaves all this diversity of habitats under a high level of threat.

Pseudochthonius ramalho sp. nov. occurs exclusively in Gruna do Vandercir cave, being considered an endemic species to its type locality. By IUCN (International Union of Conservation of Nature) criteria, we classify this species as Critically Endangered (CR) according to criteria B1ab (iii) + B2ab (iii). This means that the species has a restricted geographical distribution, with an estimated occurrence of less than 100 km^2 (B1) and 10 km^2 (B2), and the severely fragmented population (a) lives in a few locations with the continued decline (b) in area, extension, and quality of habitat (iii). Therefore, effective protection measures must be taken so that there is no degradation of this environment, which is important in several aspects, and in this case, as the limited habitat of unique species that are very sensitive to disturbances.

Acknowledgements

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) (Finance Code 001) as a scholarship to LA; PROTAX II project (Fundação de Amparo à Pesquisa do Estado de São Paulo / Fapesp 2016/50381–9 and CAPES 88887.159166/2017–00, project number 440646/2015–4), FAPESP (process 2008/05678-7 and 2010/08459–4) and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for research fellow (303715/2011–1, 308557/2014–0 and 310378/2017–6) and regular project (457413/2014–0) to MEB. We also thank: A.M.P.M. Dias, coordinator of Instituto Nacional de Ciência e Tecnologia dos Hymenoptera Parasitoides da Região Sudeste Brasileira (INCT Hympar Sudeste – FAPESP 2008/57949–4 and CNPq 573802/2008–4) for making available the equipment and L.B.R. Fernandes for taking the SEM and stereomicroscope images and for image editing; to the members of Laboratório de Estudos Subterrâneos – LES, especially J. E. Gallão and N. Hattori for the collections of specimens and help in the field trips to Serra do Ramalho. To J. E. Gallão for critical reading and suggestions to the work. To A. Gambarini for his field assistance and the images of the new species and the cave habitat. To Grupo Bambuí de Pesquisas Espeleológicas (GBPE) for sharing information about Serra do Ramalho and to all support to MEB. To Instituo Chico Mendes de Conservação da Biodiversidade (ICMBIO) for collecting permit in caves (SISBIO 20165). We thank to Mark S. Harvey and János Novák for their valuable suggestions and comments. We are also grateful to the reviewers Giulio Gardini, André Lira, one anonymous reviewer and the subject editor Martina Pavlek.

References

Andrade R (2004) Estudo populacional do pseudo-escorpião cavernícola *Maxchernes iporangae* (Chernetidae, Pseudoscorpiones). PhD thesis, São Paulo University, São Paulo.

Andrade R, Mahnert V (2003) A new cavernicolous pseudoscorpion of the genus *Spelaeobochica* Mahnert, 2001 (Pseudoscorpiones, Bochicidae) from Brazil (São Paulo State). Revue Suisse de Zoologie 110: 541–546. https://doi.org/10.5962/bhl.part.80197

Aguiar NO, Bührnheim, PF (1994) Pseudoscorpiones (Arachnida) da Bacia do Rio Urucu, Coari, Amazonas. Resumos do XX Congresso Brasileiro de Zoologia, Sociedade Brasileira de Zoologia, Rio de Janeiro.

Baptista RLC, Giupponi APL (2002) A new troglomorphic *Charinus* from Brazil (Arachnida: Amblypygi: Charinidae). Revista Ibérica de Aracnología 6: 105–110.

Beier M (1969) Ein wahrscheinlich troglobionter *Pseudochthonius* (Pseudoscorp.) aus Brasilien. Revue Suisse de Zoologie 76(1): 1–2. https://doi.org/10.5962/bhl.part.97044

Beier M (1970) Myrmecophile Pseudoskorpione aus Brasilien. Annalen des Naturhistorischen Museums in Wien 74: 51–56.

Benavides LR, Cosgrove JG, Harvey MS, Giribet G (2019) Phylogenomic interrogation resolves the backbone of the Pseudoscorpiones tree of life. Molecular Phylogenetics and Evolution 139: 106509. https://doi.org/10.1016/j.ympev.2019.05.023

Bichuette ME, Rantin B, Gallão JE (2013) A fauna subterrânea da porção sul da Serra do Ramalho. O Carste 25(1): 54–56.

Bichuette ME, Rizzato PP (2012) A new species of cave catfish from Brazil, *Trichomycterus rubbioli* sp. nov., from Serra do Ramalho karstic area, São Francisco River basin, Bahia State (Siluriformes: Trichomycteridae). Zootaxa 3480: 48–66. https://doi.org/10.11646/zootaxa.3480.1.2

Bichuette ME, Trajano E (2004) Fauna troglóbia da Serra do Ramalho, Bahia: propostas para sua conservação. O Carste 20(2): 76–81.

Bichuette ME, Trajano E (2005) A new cave species of *Rhamdia* (Siluriformes: Heptapteridae) from Serra do Ramalho, northeastern Brazil, with notes on ecology and behavior. Neotropical Ichthyology 3(4): 587–595. https://doi.org/10.1590/S1679-62252005000400016

Chamberlin JC (1929) The genus *Pseudochthonius* Balzan (Arachnida – Chelonetida). Bulletin de la Société Zoologique de France 54: 173–179.

Chamberlin JC (1931) The arachnid order Chelonethida. Stanford University Publications. Biological Sciences 7(1): 1–284.

Chamberlin JC, Malcolm DR (1960) The occurrence of false scorpions in caves with special reference to cavernicolous adaptation and to cave species in the North American fauna (Arachnida – Chelonethida). American Midland Naturalist 64: 105–115. https://doi.org/10.2307/2422895

Christiansen K (2012) Morphological adaptations. In: Culver DC, White WB (Eds) Encyclopedia of Caves, 2nd edn. Academic Press, 517–528. https://doi.org/10.1016/B978-0-12-383832-2.00075-X

Culver DC (1986) Cave Fauna. In: Soule ME (Ed.) Conservation biology: the science of scarcity and diversity. Sinauer Associates, Massachusetts, 427–443.

Faille A (2019) Beetles. In: White WB, Culver DC, Pipan T (Eds) Encyclopedia of Caves, 3rd edn. Academic Press, 102–108. https://doi.org/10.1016/B978-0-12-814124-3.00014-5

Feng Z, Wynne JJ, Zhang F (2020) Cave-dwelling pseudoscorpions of China with descriptions of four new hypogean species of *Parobisium* (Pseudoscorpiones, Neobisiidae) from Guizhou Province. Subterranean Biology 34: 61–98. https://doi.org/10.3897/subtbiol.34.49586

Fong DW (2012) General hypotheses on the mechanism of adaptation to the cave environment. In: White WB, Culver DC (Eds) Encyclopedia of Caves. Elsevier, Waltham, 341–347. https://doi.org/10.1016/B978-0-12-383832-2.00047-5

Gabbutt PD, Vachon M (1963) The external morphology and life history of the pseudoscorpion *Chthonius ischnocheles* (Hermann). Proceedings of the Zoological Society of London 140: 75–98. https://doi.org/10.1111/j.1469-7998.1963.tb01855.x

Gallão JE, Bichuette ME (2018) Brazilian obligatory subterranean fauna and threats to the hypogean environment. Zookeys 746: 1–23. https://doi.org/10.3897/zookeys.746.15140

Gao Z, Wynne JJ, Zhang F (2018) Two new species of cave-adapted pseudoscorpions (Pseudoscorpiones: Neobisiidae, Chthoniidae) from Guangxi, China. Journal of Arachnology 46(2): 345–354. https://doi.org/10.1636/JoA-S-17-063.1

Gonçalves MVP, Cruz MJM, Alencar CMM, Santos RA, Junior ABSR (2018) Geoquímica e qualidade da água subterrânea no município de Serra do Ramalho, Bahia (BR). Engenharia Sanitária e Ambiental 23(1): 159–172. https://doi.org/10.1590/s1413-41522018167893

Juberthie C, Decu V (1994) Structure et diversité du domaine souterrain: particularités des habitats et adaptations des espéces. In: Juberthie C, Decu V (Eds) Encyclopedia Biospeleologica. Société de Biospélogie, Moulis-Bucarest 1: 5–22.

Judson MLI (1992) A simple, slow–diffusion method for clearing small arthropods. The Newsletter British Arachnological Society 64: 6–7.

Judson MLI (2018) Ontogeny and evolution of the duplex trichobothria of Pseudoscorpiones (Arachnida). Zoologischer Anzeiger 273: 133–151. https://doi.org/10.1016/j.jcz.2017.12.003

Harvey MS (1992) The phylogeny and classification of the Pseudoscorpionida (Chelicerata: Arachnida). Invertebrate Taxonomy 6: 1373–1435. https://doi.org/10.1071/IT9921373

Harvey MS (2013) Pseudoscorpions of the World. Version 3.0. Western Australian Museum, Perth. http://www.museum.wa.gov.au/catalogues–beta/pseudoscorpions [accessed 21 July 2020]

Harvey MS (2021) A new genus of the pseudoscorpion family Chernetidae (Pseudoscorpiones) from southern Australia with Gondwanan affinities. Journal of Arachnology 48: 300–310. https://doi.org/10.1636/JoA-S-20-038

Harvey MS, Cullen KL (2020) A remarkable new troglobitic *Parobisium* (Pseudoscorpiones: Neobisiidae) from California. Arachnology 18: 591–596. https://doi.org/10.13156/arac.2020.18.6.591

Harvey MS, Wynne JJ (2014) Troglomorphic Pseudoscorpions (Arachnida: Pseudoscorpiones) of Northern Arizona, with the Description of Two New Shortrange Endemic Species. The Journal of Arachnology 42: 205–219. https://doi.org/10.1636/K14-34.1

Harvey MS, Shear WA, Hoch H (2000) Onychophora, Arachnida, Myriapods and Insecta. In: Wilkens H, Culver DC, Humphreys WF (Eds) Ecosystems of the World: Subterranean Ecosystems. Elsevier, 79–94.

Hertault J (1994) Pseudoscorpions. In: Juberthie C, Decu V (Eds) Encyclopaedia Biospeologica. Société de Biospélogie, Moulis and Bucarest 1: 185–196.

Lira AFA, Bedoya-Roqueme E, Rodrigues GG, Tizo-Pedroso E (2020) New records of pseudoscorpions (Arachnida, Pseudoscorpiones) from the Caatinga biome, Brazil: a checklist and a map of species richness distribution. CheckList 16(2): 471–484. https://doi.org/10.15560/16.2.471

Mahnert V (1979) Pseudoskorpione (Arachnida) aus dem Amazonas-Gebiet (Brasilien). Revue Suisse de Zoologie 86: 719–810. https://doi.org/10.5962/bhl.part.82338

Mahnert V (2001) Cave-dwelling pseudoscorpiones (Arachnida, Pseudoscorpiones) from Brazil. Revue Suisse de Zoologie 108: 95–148. https://doi.org/10.5962/bhl.part.79622

Mahnert V, Sharaf M, Aldawood AS (2014) Further records of pseudoscorpions (Arachnida, Pseudoscorpiones) from Saudi Arabia. Zootaxa 3764(3): 387–393. https://doi.org/10.11646/zootaxa.3764.3.8

Mahnert V, Adis J (2002) Pseudoscorpiones. In: Adis J (Ed.) Amazonian Arachnida and Myriapods. Pensoft Publishers, Sofia, 367–380.

Muchmore WB (1970) An unusual new *Pseudochthonius* from Brazil (Arachnida, Pseudoscorpionida, Chthoniidae). Entomological News 81: 221–223.

Muchmore WB (1986) Additional pseudoscorpions, mostly from caves, in Mexico and Texas (Arachnida: Pseudoscorpionida). Texas Memorial Museum. Speleological Monograph 1: 17–30.

Pérez AG, Kury AB (2002) A new remarkable troglomorphic Gonyleptid from Brazil (Arachnida, Opiliones, Laniatores). Revista Ibérica de Aracnología 5: 43–50.

Ratton P, Mahnert V, Ferreira RL (2012) A new cave-dwelling species of *Spelaeobochica* (Pseudoscorpiones: Bochicidae) from Brazil.Journal of Arachnology 40: 274–280. https://doi.org/10.1636/Ha12-39.1

Reddell JR (2012) Spiders and related groups. In: White WB, Culver DC (Eds) Encyclopedia of Caves. Elsevier, Waltham, 786–797. https://doi.org/10.1016/B978-0-12-383832-2.00114-6

Rubbioli E, Auler A, Menin D, Brandi R (2019) Cavernas – Atlas do Brasil Subterrâneo. Instituto Chico Mendes da Biodiversidade, Brasília, 340 pp.

Schimonsky DM, Bichuette ME, Mahnert V (2014) First record of the family Pseudochiridiidae (Arachnida, Pseudoscorpiones) from continental South America – a *Pseudochiridium* from a Brazilian cave. Zootaxa 3889(3): 442–446. https://doi.org/10.11646/zootaxa.3889.3.6

Schimonsky DM, Bichuette ME (2019a) A new cave–dwelling *Spelaeochernes* (Pseudoscorpiones: Chernetidae) from northeastern Brazil. Journal of Arachnology 47(2): 248–259. https://doi.org/10.1636/JoA-S-16-086

Schimonsky DM, Bichuette ME (2019b) Distribution of cave–dwelling pseudoscorpions (Arachnida) in Brazil. Journal of Arachnology 47(1): 110–123. https://doi.org/10.1636/0161-8202-47.1.110

Silva Junior AF, Campos MF (2016) Relevance of rare earth for the energy sector. Holos 32(1): 350–363. https://doi.org/10.15628/holos.2016.3753

Trajano E, Secutti S, Bichuette ME (2009) Natural history and population data of fishes in caves of the Serra do Ramalho karst área, Middle São Francisco basin, northeastern Brazil. Biota Neotropica 9(1): 129–133. https://doi.org/10.1590/S1676-06032009000100015

Viana ACM, Souza MFVR, Ferreira RL (2018) *Spelaeobochica goliath* (Arachnida: Pseudoscorpiones: Bochicidae), a new troglobitic pseudoscorpion from Brazil. Zootaxa 4402(3): 585–594. https://doi.org/10.11646/zootaxa.4402.3.11

Viana ACM, Ferreira RL (2020) *Spelaeobochica mahnerti*, a new cave–dwelling pseudoscorpion from Brazil (Arachnida: Pseudoscorpiones: Bochicidae), with comments on the troglomorphism of the Brazilian bochicid species. Zootaxa 4731(1): 134–144. https://doi.org/10.11646/zootaxa.4731.1.9

Wagenaar-Hummelinck P (1948) Studies on the fauna of Curaçao, Aruba, Bonaireand the Venezuelan Islands: No. 13. Pseudoscorpions of the genera *Garypus*, *Pseudochthonius*, *Tyrannochthonius* and *Pachychitra*. Natuurwetenschappelijke Studiekring voor Suriname en Curaçao 5: 29–77.

Zhang F, Zhang F (2014) First report of the family Lechytiidae (Arachnida: Pseudoscorpiones) from China, with the description of a new species. Acta Zoologica Academiae Scientiarum Hungaricae 60(3): 217–225.

Zaragoza JA, Reboleira ASPS (2018) Five new hypogean *Occidenchthonius* (Pseudoscorpiones: Chthoniidae) from Portugal. Journal of Arachnology 46(1): 81–103. https://doi.org/10.1636/JoA-S-17-031.1